MARKED-UP VERSION OF AMENDMENTS

The specification has been amended to replace commas by periods in number notation (American scientific notation). Further, the term "concentrations" has been changed to "doses" on page 32, line 21, and the terms "Intact (norm)" has been changed to "Norm" and the term "Together" has been changed to "Intact" in Table 11 on page 47 and Table 13 on page 49.

Claims 1-5 and 20 have been amended as follows:

1. (Amended) A method for the prevention or treatment of inflammation or inflammatory-related disorder comprising administering to a mammal in need of such treatment a composition comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent, said composition comprising said ribonucleic acid in an amount effective to ameliorate [the] symptoms of inflammation or inflammatory-related disorder [of ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent], wherein said composition is administered so that said ribonucleic acid is present into the mammal's blood.

2. (Amended) A method of stabilizing [damaged cellular] acid-challenged erythrocyte membranes of a mammal in need of prevention or treatment of inflammation or an inflammatory-related disorder, which comprises administering to [a] said mammal [having damaged cellular membranes] a composition comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent, said composition containing said total yeast ribonucleic acid in an amount effective to stabilize said [damaged cellular] acid-challenged erythrocyte membranes [of ribonucleic

acid and a pharmaceutically acceptable vehicle, carrier, or diluent], wherein said composition is administered so that said ribonucleic acid is present into the mammal's blood.

3. (Amended) A method of inhibiting oxidation into arachidonic acid of components of cell membranes of a mammal in need of prevention or treatment of inflammation or an inflammatory-related disorder, which comprises administering to [a] said mammal [in need of such treatment] a composition comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent, said composition containing said total yeast ribonucleic acid in an amount effective to inhibit oxidation into arachidonic acid of components of cell membranes of the mammal [of ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent], wherein said composition is administered so that said ribonucleic acid is present into the mammal's blood.

4. (Amended) A method of [normalization of] reducing an amplitude of variations of NO-synthetase activity induced by inflammation or inflammatory-related disorder in a mammal, which comprises administering to [a] said mammal [in need of such theatment] a composition comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent, said composition containing said total yeast ribonucleic acid in an amount effective to [normalize] reduce the amplitude of the variations of NO-synthetase activity in the mammal [of ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent], wherein said composition is administered so that said ribonucleic acid is present into the mammal's blood.

5. (Amended) A method of inhibiting thrombocyte aggregation induced by inflammation or inflammatory-related disorder, which comprises administering to a mammal in need of such treatment a composition comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle,

carrier, or diluent, said composition containing said total yeast ribonucleic acid in an amount effective to inhibit thrombocyte aggregation [of ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent], wherein said composition is administered so that said ribonucleic acid is present into the mammal's blood.

20. (Amended) A pharmaceutical composition for the treatment or the prevention of inflammation or inflammatory-related disorder, comprising total yeast ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent.

New claims 23-38 have been added.

REMARKS

By the present amendment, claims 1-5 and 20 have been amended and claims 23-38 have been added.

Claims 1-38 are pending in the present application. Claims 1-19 and 23-37 are directed to methods for the treatment of various affections comprising administration of ribonucleic acid, and claims 20-22 and 38 are directed to pharmaceutical compositions comprising ribonucleic acid.

I. INTRODUCTION

As a preliminary, the Applicant submits that the anti-inflammatory action of total yeast ribonucleic acid (RNA) has been demonstrated in the present specification using scientifically acknowledged models, and in particular:

- thrombocytic aggregation under the influence of arachidonic acid and carageenan-induced swelling, which identified total yeast RNA as a very effective anti-inflammation agent;
- acid resistance of erythrocytic membranes, both in vitro and in vivo, which allowed the identification of anti-inflammatory action of total yeast RNA on the membrane as main target;
- NOS activity, which showed effectiveness of total yeast RNA at early stages of inflammation;
- adjuvant arthritis, which illustrated the membrane stabilization activity and long-term effect of total yeast RNA;
- ischemia reperfusion, which illustrated inhibition of enzyme oxidation in cell membranes by total yeast RNA.

Further, the Applicant submits that these models for inflammatory processes, as used in the examples of the present specification, have been recognized as valid scientifically and pharmacologically. Thus, these models are scientifically valid models for predicting general or specific anti-inflammatory effects of a composition.

In order to illustrate this point, reference is made to exemplary documents selected among scientific literature regarding these inflammation models. For example, Yukio Sato et al., Mechanism of Free Radical-Induced Hemolysis of Human Erythrocytes: Hemolysis by Water-Soluble Radical Initiator, Biochem., 1995, 34, 8940-8949, reports that "the oxidation of erythrocyte or its ghost membranes by free radicals has been used as a model for the oxidative damages of biomembranes" (page 8940, left column, second paragraph). Koichi Katsuyama et al., Differential Inhibitory Actions by Glucocorticoid and Aspirin on Cytokine-Induced Nitric Oxide Production in Vascular Smooth Muscle Cells, Endocrinology, 1999, 140, 2183-2190, uses a model of induced NO production in muscle cells. R. Walter et al., Differential Regulation of Constitutive and Inducible Nitric Oxide Production by Inflammatory Stimuli in Murine Endothelial Cells, Biomedical and Biophysical Research Communications, 1994, 202 (1), 450-455, and John R. Vane, Inducible Isoforms of Cyclooxygenase and Nitric-Oxide Synthase in Inflammation, Proc. Natl. Acad. Sci USA, 1994, 91, 2046-2050, also use NO production models. Jarle Vaage et al., Myocardial Reperfusion Injury: an Inflammation Reaction?, Biomed. Biochim. Acta 48, 1989, 2/3, 63-68, uses "models of surgically induced, hypothermic, global ischemia" with rats to study ischemia (page 64, second full paragraph). A copy of each of these documents is enclosed.

See also, e.g., U.S. Patents Nos. US 4,192,891 to Haslanger (arachidonic acid-induced aggregation model), US 5,321,041 to Adachi et al., US 5,449,783 to Saita et al., US 6,221,887 to Asghar et al., US 6,274,177 to Wu et al. (Carageenan and thrombocytic aggregation models).

The objections and rejections set forth in the Office Action will now be addressed.

I. OBJECTION TO THE SPECIFICATION

On page 2 of the Office Action, the specification is objected to. It is requested that errors and lack of precision in the language and scientific notations be corrected.

The specification has been reviewed and obvious errors have been corrected. In particular, scientific notation has been uniformized throughout the specification, on page 32, line 21, the term "concentrations" has been replaced by "doses" and in Tables 11 and 13, the term "Together" has been replaced by "Intact." Also,.

II. INDEFINITENESS REJECTION

Next, on pages 2-4 of the Office Action, claims 1-19 are rejected under 35 U.S.C. 112, second paragraph, as indefinite. It is alleged that, in claims 1-5, the recitations "A method for the treatment of... comprising administering to a mammal... an amount effective to [ameliorate..., stabilize..., inhibit..., normalize..., inhibit..., respectively] of ribonucleic acid..." does not clarify what is being administered, and that, in claim 4, the recitation "normalize" is unclear because it does clarify the circumstances of an abnormality in the enzymatic mechanism in vivo.

Reconsideration and withdrawal of the rejection is respectfully requested. Claims 1-5 have been amended to use the recitation "A method for the treatment of... comprising administering to a mammal in need of such treatment a composition comprising ribonucleic acid and a pharmaceutically acceptable vehicle, carrier, or diluent, said composition containing ribonucleic acid in an amount effective to..." which clarifies that the composition is being administered.

Further, with respect to claim 4, reference is made to the passage on pages 22-23 of the specification which explains the correlation between inflammation and NOS activity in vivo, and describes the conventional carrageenan-induced model which is used in Example 4.2.1. on pages 36-39. Thus, as explained and illustrated in the specification, inflammatory processes involve a drastic modification of NOS activity, which may be a rapid increase (as happens to constitutive NOS at earlier stages, when inflammation mediators are turned on) or a rapid decrease (as happens to inductive NOS at later stages in a tumor process, especially in chronic inflammations), or a wave-like fluctuation with a large amplitude. The method of claim 4 includes regulating the activity of NOS.

In order to clarify this point, claim 4 has been amended to replace the terms "normalization of NO-synthetase activity" and "normalize NO-synthetase activity" by "reducing the amplitude of NO-synthetase activity" and "reduce the amplitude of NO-synthetase activity."

In view of the above, it is submitted that the indefiniteness rejection should be withdrawn.

III. LACK OF ENABLEMENT REJECTION

Next, on pages 5-12 of the Office Action, claims 1-4 and 6-19 are rejected under 35 U.S.C. 112, first paragraph, as not enabled. In summary, it is alleged in the Office Action that the description

of the invention set forth in the application is only enabling for (i) short term effects, (ii) using yeast RNA, (iii) which is injected interabdominally, (iv) prior to the inflammatory event.

In contrast, it is alleged in the Office Action that the description is insufficient for effects on chronic inflammatory diseases, using total RNA extracted from other sources than yeast, with other routes of administration. In particular, it is alleged:

- that previously known methods using RNA involved specific RNA sequences with specific cellular targets and action mechanisms, so that action of total RNA from an organism to another is unpredictable to a person of the art, that oral or hypodermal administration would be expected to cause degradation of RNA, and that the experimental data in the specification only support short term effect, for example on NOS;
- with respect to claim 2, that the specification is enabling only for the specification *in vitro* conditions of the examples (stabilization of acid challenged erythrocyte plasma membrane isolated from carageenan treated rats using total RNA from S cerevisiae which is interabdominally injected), and that cells other than erythrocytes have membrane components different from erythrocytes which are specialized cells, and that inflammation processes include membrane destabilization processes different from acid lysis;
- with respect to claim 3, similarly, that the specification is enabling only for inhibition of phospholipid oxidation producing arachidonic acid, using total RNA from *S. cerviseae* which is interabdominally injected to a rat or mouse ;
- with respect to claim 4, that the data in the specification is contradictory and does not unequivocally show normalization of NOS activity levels.

Reconsideration and withdrawal of the rejection is respectfully requested.

As a preliminary, claims 1-5 have been amended to recite that the RNA is total yeast RNA and that the RNA is administered into the blood of the mammal. It is submitted that a person of the art would immediately understand that the anti-inflammatory effect of RNA is obtained when RNA is present in the blood of the mammal, for example as a result of interabdominal injection as illustrated in the examples.

However, the Applicant respectfully traverses the position set forth in the Office Action that the specification is enabling only when RNA is administered prior to occurrence of the inflammation. It is submitted that various examples in the present specification include RNA administration during or prior to the development of inflammatory reactions. For example, in Example 6, RNA is administered over 14 days after onset of inflammation. Thus, a person of the art would understand that RNA administration can be effective when administered before, during or after onset of inflammatory processes. As a result, the present specification is enabling for such RNA administration.

injected before disease development

The Applicants also traverse the position set forth in the Office Action that the specification is enabling only for "short term" effects. It is submitted that an important factor in the interpretation of inflammatory processes is not the time or duration, but the presence of the various representative stages of the inflammatory reaction. Thus, various examples in the present specification consider short term effects in the case of inflammatory processes which develop in a short timeframe. For example, the experiments reported in the present specification involve short periods of time (30, 60 and 320 minutes) when NOS activity and AA degradation processes are considered, as required by

the accelerated evolution of such processes. Conversely, other examples in the present specification are directed to long-term effects of the methods according to the presently claimed invention. Thus, Example 6 shows that the preventive or treating action of yeast RNA is shown over periods of up to 14 days (Example 6.2) and 30 days (Example 6.1). The examples in the present specification were selected to use appropriate inflammatory reaction models. Thus, some models are such that all stages of an inflammatory reaction are complete in 4 hours (e.g., carageenan model) while others involve inflammatory processes which develop over up to 14 days (e.g., adjuvant arthritis model).

Specifically, with respect to Table 6 on page 37 of the present specification, it is submitted that this Table illustrates that RNA is effective against constitutive NOS at the earliest stage of inflammation, i.e., 30-60 minutes. Thus, RNA has a strong inhibiting effect against constitutive NOS at the early stage of the inflammatory process. Since constitutive NOS, along with other signal systems, leads to the activation of cells which secrete inflammatory mediators, RNA reduces the extent of the later stages as well. In particular, RNA is considerably more effective than aspirin which is effective only against inductive NOS, and thus, not active at the earliest stages of inflammation, but only at the less important later stages of the inflammatory processes, when production of inflammatory mediators have already been activated. In short, the experimental results reported on Table 6 confirm that RNA has both preventive and therapeutic effects by inhibiting NOS activity at the early stage of inflammation.

The Applicant wishes to point out again that the models for inflammatory processes (i.e., damage to cellular membranes, membrane oxidation, NOS activity, thrombocyte aggregation) which are used in the examples of the present specification have been recognized as valid scientifically and

pharmacologically, so that these models are scientifically valid models for predicting the anti-inflammatory effect of a composition.

In particular, it is submitted the various experimental models which are used in the present specification are predictive of results, not only for mice and rats, but for other mammals as well, as recognized and accepted by the scientific community. In addition, reference is made to Example 2 which uses thrombocytes obtained from venous human blood in a scientifically recognized model of thrombocytic aggregation under the influence of arachidonic acid. Therefore, the present specification establishes and illustrates the anti-inflammatory action of total yeast RNA, not only for mice and rats, but for other mammals, including humans, as well.

Specifically, the carageenan model is known in the art and predictive of other inflammations. Reference is made in particular to Example 6.3 which provides an erythrocyte membrane resistance model for an auto-immune process (adjuvant arthritis). Thus, with respect to claim 2, the present specification is enabling for damaged membrane stabilization, not only for caraagenan-induced inflammations and adjuvant arthritis inflammations, but for other types of inflammations as well.

In summary, the method of the present invention as claimed in present claims 1-5 is enabled in the present specification.

In addition, with respect to claims 23-24, which are dependent on claim 1, it is submitted that claim 23 recites interabdominal injection, and that claim 24 recites short term effect. Therefore, for this reason alone, claims 23-24 are enabled.

With respect to present claims 25-28, which are dependent on claim 2, and recite inflammatory swelling which may be carageenan-induced, and auto-immune inflammation which may

be adjuvant arthritis, respectively, it is submitted that these claims are enabled in the present specification.

With respect to new claims 29-32, which are dependent on claims 2-5, respectively, and recite that the mammal is a rat or mouse, it is submitted that these claims are enabled in the present specification.

With respect to new claims 33-37, which are dependent on claims 1-5, respectively, and recite that the composition is administered so that RNA is present into the mammal's blood prior to the condition, it is submitted that these claims are enabled in the present specification.

In view of the above, it is submitted that the lack of enablement rejection should be withdrawn.

IV. PRIOR ART REJECTIONS

Next, on pages 13-16 of the Office Action, the following prior art rejections are set forth:

- claims 1, 6, 10-12 and 14-22 are rejected under 35 U.S.C. 102(b) as anticipated by WO94/02595 (**Sullivan**) (pages 13-14 of the Office Action) and claims 2-5 are rejected under 35 U.S.C. 103(a) as obvious over **Sullivan** (pages 15-16 of the Office Action); and
- claims 20-22 are rejected under 35 U.S.C. 102(b) as anticipated by Tait et al., Clinical Cancer Research (1997) 3:1959-1967 (**Tait**) (pages 14-15 of the Office Action).

Reconsideration and withdrawal of the rejections is respectfully requested. Both **Sullivan** and **Tait** refer to administration of specific RNA sequences which are not originated from yeast. Therefore, these references fail to teach or suggest administration of total yeast RNA as recited in

the present claims. Specifically, the RNA of **Sullivan** corresponds to ribozymes which are targeted at the destruction of specific mRNA transcripts. In contrast, yeast RNA lacks this particular activity. Further, **Tait** describes retroviral insertion of a specific BRCA1sv gene, which involves very complicated gene therapy. In contrast, yeast RNA is active through different processes and can be administered considerably more easily. Thus, the features of the presently claimed invention and their advantages are not taught or suggested in any of the cited references. As a result, the present claims are not obvious over the cited references taken alone or in any combination.

In view of the above, it is submitted that the prior art rejection should be withdrawn.

V. CONCLUSION

In conclusion, the invention as presently claimed is patentable. It is believed that the claims are in allowable condition and a notice to that effect is earnestly requested.

In the event there is, in the Examiner's opinion, any outstanding issue and such issue may be resolved by means of a telephone interview, the Examiner is respectfully requested to contact the undersigned attorney at the telephone number listed below.

In the event this paper is not considered to be timely filed, the Applicants hereby petition for an appropriate extension of the response period. Please charge the fee for such extension and any other fees which may be required to our Deposit Account No. 01-2340.

Respectfully submitted,

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- Yukio Sato et al., Mechanism of Free Radical-Induced Hemolysis of Human Erythrocytes: Hemolysis by Water-Soluble Radical Initiator, Biochem., 1995, 34, 8940-8949,
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